

# HIGH ABUNDANCE OF NONTUBERCULOUS MYCOBACTERIA IN HOUSEHOLDS AND URBANIZED ENVIRONMENTS IN POLAND

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## BACKGROUND

Due to the rise in the number of documented non-tuberculous mycobacteria (NTM) cases along with people at higher risk, NTM have been declared an emerging public health threat and earned a status of “the new uber-bugs”. However, the definite ecological niches for most of the NTM remain poorly understood. The purpose of this study was to examine the occurrence of NTM in households and urbanized environments in Poland using conventional culture-based methods.

## MATERIALS & METHODS

A total of 156 samples were collected from households and urbanized environments in Poland between April and August 2023. The household samples (n=103) included tap water (n=26), pipe sludge (n=25), tap biofilm (n=26), and air (n=26). The environmental samples (n=53) consisted of soil (n=12), air (n=21) and different water sources (n=20). Samples were decontaminated and cultivated on NTM Elite agar and Middlebrook 7H11 supplemented with glycerol, OADC and 25 mg/L malachite green. The incubation was carried out at 30°C for 6 weeks in the dark. Acid-fast bacilli (AFB+) were identified to the species level by PCR-sequencing of the *hsp65* gene (Fig. 1).

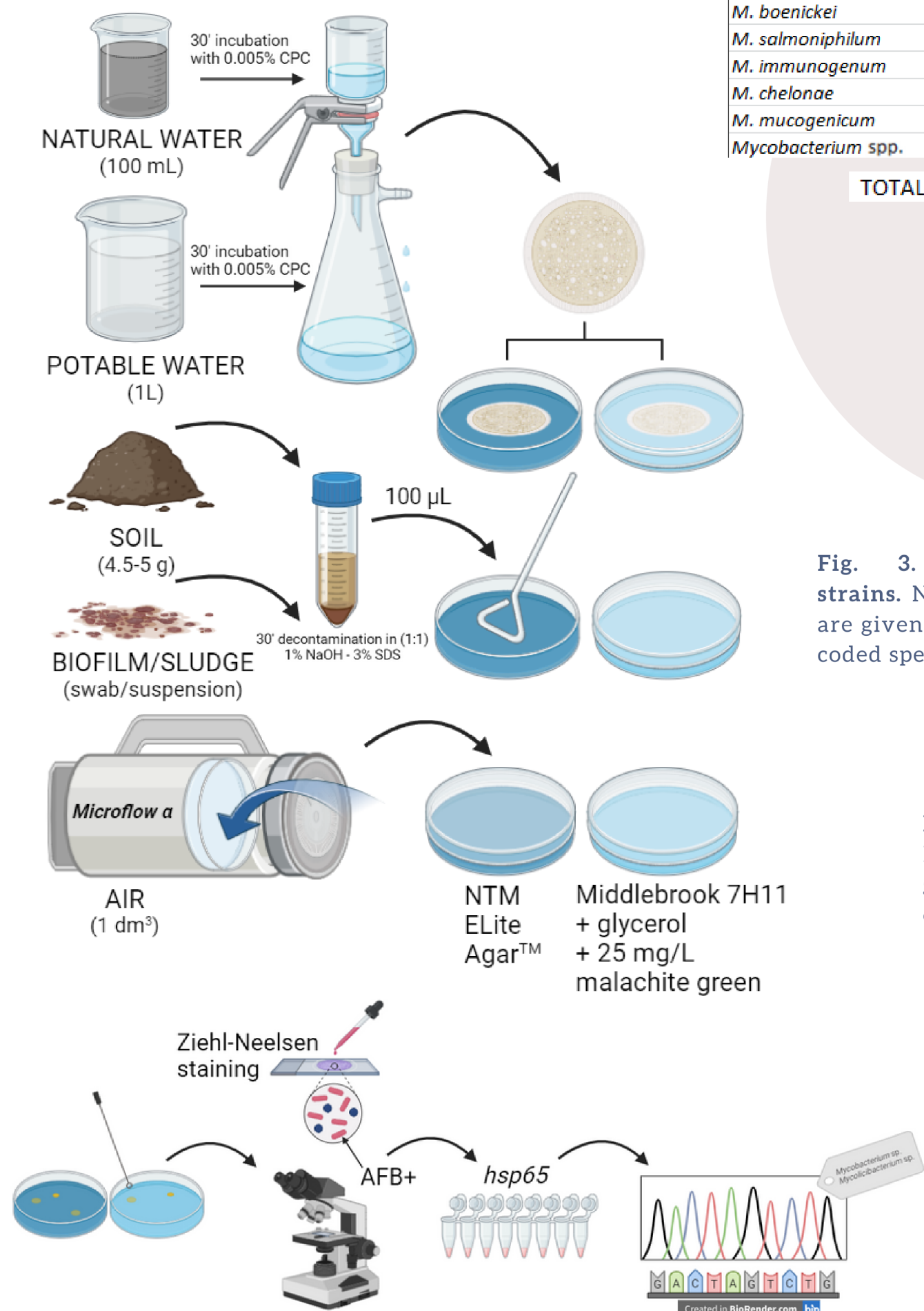


Fig. 1. Methodology. Collected samples were decontaminated and cultured on two media promoting NTM growth. Acquired strains were identified by PCR-seq.

## RESULTS

Of the 156 samples collected, 91 (58.3%) yielded mycobacterial growth. In this number there were 68 (66%) household and 23 (44.2%) environmental samples. Among household sources, the most NTM-abundant was air (88%), followed by tap water (77%), tap biofilm (52%), and pipe sludge (46%). As for the environmental samples, the highest ratio of isolated NTM was observed for fountains (100%), followed by soil (66.7%), flowing water (42.9%), air (33.3%), and stagnant water (25%).

More than 30 different species of mycobacteria were identified. Most isolates (63.4%) were described to the species level, meeting the criteria of at least 98% *hsp65* sequence identity (BLASTn). The most frequently occurring household species (Fig.2) was *M. gordonae* followed by *M. mucogenicum*. Among environmental samples (Fig.3), the most common NTM were *M. chelonae* and *M. mucogenicum*, followed by *M. immunogenum*.

<i>M. fluoranthenorans</i>	1
<i>M. peregrinum</i>	1
<i>M. septicum</i>	1
<i>M. llatzerense</i>	1
<i>M. avium</i>	1
<i>M. gordonae</i>	1
<i>M. insubricum</i>	2
<i>M. setense</i>	2
<i>M. boenickei</i>	3
<i>M. salmoniphilum</i>	3
<i>M. immunogenum</i>	4
<i>M. chelonae</i>	5
<i>M. mucogenicum</i>	5
<i>Mycobacterium</i> spp.	26

TOTAL: 56

<i>M. arceuilense</i>	1
<i>M. asiaticum</i>	1
<i>M. gilvum</i>	1
<i>M. heidelbergense</i>	1
<i>M. hiberniae</i>	1
<i>M. intracellulare</i>	1
<i>M. iranica</i>	1
<i>M. novocastrense</i>	1
<i>M. paraense</i>	1
<i>M. parafortuitum</i>	1

<i>M. phlei</i>	1
<i>M. porcinum</i>	1
<i>M. immunogenum</i>	1
<i>M. fortuitum</i>	2
<i>M. nivoideus</i>	2
<i>M. nonchromogenicus</i>	2
<i>M. boenickei</i>	2
<i>M. llatzerense</i>	3

<i>M. lentiflavum</i>	4
<i>M. phocaicum</i>	4
<i>M. paraintracellulare</i>	5
<i>M. avium</i>	6
<i>M. chelonae</i>	9
<i>M. salmoniphilum</i>	10
<i>M. paragordonae</i>	13
<i>M. mucogenicum</i>	14
<i>M. gordonae</i>	18
<i>Mycobacterium</i> spp.	60

TOTAL: 167

Fig. 3. Environmental strains. Number of isolates are given next to the color-coded species.

Fig. 2. Household strains. Number of isolates are given next to the color-coded species.

## CONCLUSIONS

- This is the first investigation into the distribution of NTM in natural and human-engineered habitats in Poland;
- NTM were highly abundant in both natural and anthropogenic environments;
- NTM species distribution differed between household and environmental samples;
- M. avium*, *M. boenickei*, *M. chelonae*, *M. gordonae*, *M. immunogenum*, *M. llatzerense*, *M. mucogenicum*, and *M. salmoniphilum* were present in both environmental and household samples.
- Pathogenic NTM species were identified in the analyzed samples, including air and tap water.